

What is claimed is:

1. A method to detect expression of HAS isoenzyme variants in a cell or cell population comprising contacting an agent capable of selectively binding to HAS isoenzyme variant genomic products with a sample of cell or cell population, then detecting the presence of complex formed between said agent and the HAS isoenzyme variant genomic product.
2. The method of claim 1 wherein the isoenzyme variant genomic product is a nucleotide.
3. The method of claim 2 wherein the nucleotide is mRNA.
4. The method of claim 2 wherein said nucleotide is generated using cellular mRNA as a template.
5. The method of claim 3 wherein the agent capable of selectively binding to HAS isoenzyme variant mRNA is a nucleotide.
6. The method of claim 2, 3, 4, or 5 wherein the presence of the complex formed is detected using a fluorescent label.
7. The method of claim 4 wherein the nucleotide produced using mRNA as a template is cDNA.
8. The method of claim 7 wherein the agent capable of selectively binding to HAS isoenzyme variant cDNA is a nucleotide.
9. The method of claim 7 wherein the presence of the complex formed is detected using a fluorescent label.
10. The method of claim 1 wherein the method is performed using a microfluidic device.

11. The method of claim 1 wherein the HAS isoenzyme variant genomic product is a polypeptide.
12. The method of claim 11 wherein the agent capable of selectively binding to the polypeptide HAS isoenzyme variant genomic product is an antibody.
- 5 13. The method of claim 11 wherein the agent capable of selectively binding to the polypeptide HAS isoenzyme variant genomic product is an antibody fragment.
14. The method of claim 12 or 13 wherein the complex formed is detected using a fluorescent label.
15. The method of claim 12 or 13 wherein the complex formed is detected using a radiolabel.
- 10 16. The method of claim 12 or 13 wherein the complex formed is detected using a colorimetric reaction.
17. The method of claim 1, 2, or 11 wherein the HAS isoenzyme variant is HAS1Va.
18. The method of claim 1, 2, or 11 wherein the HAS isoenzyme variant is HAS1Vb.
- 15 19. The method of claim 1, 2, or 11 wherein the HAS isoenzyme variant is HAS1Vc.
20. The method of claim 1, 2, or 11 wherein the HAS isoenzyme is HAS2.
21. A method to detect expression of HAS1 isoenzyme variants comprising:
- i) mixing a cell or sample of cell population from a human with reverse transcriptase in conditions enabling conversion of mRNA to DNA templates thereby generating cDNA templates;
- 20 ii) mixing said cDNA with at least two oligonucleotide primers specific for HAS1, whereby primers are selected so as to enable generation of amplified fragments of

differing size for each HAS1 isoenzyme variant;

a. Reacting said mixture with enzymes and compounds to enable specific fragments of DNA to be increased in number;

b. Detecting the presence of an increased number of resulting DNA fragments of particular size associated with the presence of particular HAS1 isoenzyme variants.

22. The method of claim 21 wherein the oligonucleotide primers selected are SEQ ID NO: 9 and SEQ ID NO:10

23. The method of claim 21 wherein the isoenzyme variant is HAS1Va.

24. The method of claim 21 wherein the isoenzyme variant is HAS1Vb.

25. The method of claim 21 wherein the isoenzyme variant is HAS1Vc.

26. The method of claim 21 wherein the isoenzyme is HAS2.

27. The method of claim 21 wherein the process is performed using a microfluidic device.

28. A method to detect expression of HAS1Va isoenzyme variant in a cell or cell population comprising detection of single nucleotide polymorphism of the HAS1Va gene.

29. The method of claim 28 wherein the single nucleotide polymorphism results in conversion of base 924 of the HAS1Va cDNA from a cytosine to a thymidine residue.

30. A method to detect disease comprising characterizing HAS isoenzyme and isoenzyme variant expression in a cell or cell population.

31. The method of claim 30 wherein the disease results from genetic instability.

32. The method of claim 30 wherein characterizing of HAS isoenzyme and isoenzyme

variants comprises detection of HAS1 isoenzyme variants.

33. The method of claim 30 wherein the cell or cell population is selected from the group comprising blood, B-cells, CD 19<sup>+</sup> B cells, CD 19<sup>+</sup> peripheral blood mononuclear cells and bone marrow plasma cells.

5 34. The method of claim 29 or 33 wherein the characterization of HAS isoenzyme variant expression comprises detection of HAS1Va.

35. The method of claim 29 or 33 wherein the characterization of HAS isoenzyme variant expression comprises detection of HAS1Vb.

36. The method of claim 29 or 33 wherein the characterization of HAS isoenzyme variant expression comprises detection of HAS1Vc.

37. The method of claim 29 or 33 wherein the characterization of HAS isoenzyme expression comprises detection of HAS2.

38. A method to detect susceptibility to disease comprising characterizing of HAS isoenzyme or isoenzyme variant expression in a cell or cell population.

15 39. The method of claim 38 wherein characterization of HAS isoenzyme and isoenzyme variants comprises detection of HAS1 isoenzyme variants

40. The method of claim 38 wherein the cell or cell population is selected from the group comprising blood, B-cells, CD 19<sup>+</sup> B cells, CD 19<sup>+</sup> peripheral blood mononuclear cells and bone marrow plasma cells.

20 41. The method of claim 40 wherein the characterization of HAS isoenzyme variant expression comprises detection of HAS1Va.

42. The method of claim 40 wherein the characterization of HAS isoenzyme variant

expression comprises detection of HAS1Vb.

43. The method of claim 40 wherein the characterization of HAS isoenzyme variant expression comprises detection of HAS1Vc.

44. The method of claim 40 wherein the characterization of HAS isoenzyme expression comprises detection of HAS2.

45. The method of claim 38 wherein the susceptibility to disease results from genetic instability.

46. A method to detect susceptibility to disease comprising detection of single nucleotide polymorphism within the HAS1Va gene.

47. The method of claim 46 wherein the single nucleotide polymorphism within the HAS1Va gene is detected within the mRNA of a cell or cell population.

48. The method of claim 46 wherein the single nucleotide polymorphism within the HAS1Va gene is detected within cDNA generated from the mRNA of a cell or cell population.

49. A method to determine the likelihood of poor clinical outcome in a human suffering from multiple myeloma comprising characterizing HAS isoenzyme or isoenzyme variant expression in a cell or cell population.

50. The method of claim 49 wherein the cell or cell population is selected from the group comprising blood, B-cells, CD 19<sup>+</sup> B cells, CD 19<sup>+</sup> peripheral blood mononuclear cells and bone marrow plasma cells.

51. An isolated and purified DNA molecule comprising a DNA segment encoding a human hyaluronan synthase variant or enzymatically active fragment thereof, wherein

the DNA molecule hybridizes under stringent conditions to SEQ ID NO:3 or complement thereof.

52. The DNA molecule of claim 51 wherein the DNA segment encodes the human hyaluronan synthase isoenzyme variant HAS1Va.

5 53. The DNA molecule of claim 51 wherein the DNA segment encodes a hyaluronan synthase having SEQ ID NO: 4.

54. An isolated and purified DNA molecule comprising a DNA segment encoding a human hyaluronan synthase variant or enzymatically active fragment thereof, wherein the DNA molecule hybridizes under stringent conditions to SEQ ID NO:5 or  
10 complement thereof.

55. The DNA molecule of claim 54 wherein the DNA segment encodes the human hyaluronan synthase isoenzyme variant HAS1Vb.

56. The DNA molecule of claim 54 wherein the DNA segment encodes a hyaluronan synthase having SEQ ID NO: 6.

15 57. An isolated and purified DNA molecule comprising a DNA segment encoding a human hyaluronan synthase variant or enzymatically active fragment thereof, wherein the DNA molecule hybridizes under stringent conditions to SEQ ID NO:7 or complement thereof.

58. The DNA molecule of claim 57 wherein the DNA segment encodes the human  
20 hyaluronan synthase isoenzyme variant HAS1Vc.

59. The DNA molecule of claim 57 wherein the DNA segment encodes a hyaluronan synthase having SEQ ID NO: 8.

60. An isolated and purified DNA molecule comprising a DNA segment capable of selectively binding to the mRNA of human hyaluronan synthase isoenzyme-1 (HAS1) or nucleotide product thereof; allowing, when used in conjunction with a corresponding downstream DNA segment capable of selectively binding to mRNA of human hyaluronan synthase isoenzyme-1 (HAS1) or nucleotide product thereof; DNA fragment amplification and identification of HAS1 isoenzyme variants, wherein the DNA molecule hybridizes under stringent conditions to SEQ ID NO:9.
61. An isolated and purified DNA molecule comprising a DNA segment capable of selectively binding to the mRNA of human hyaluronan synthase isoenzyme-1 (HAS1), or nucleotide product thereof; allowing, when used in conjunction with a corresponding downstream DNA segment capable of selectively binding to mRNA of human hyaluronan synthase isoenzyme-1 (HAS1), or nucleotide product thereof; DNA fragment amplification and identification of HAS1 isoenzyme variants, wherein the DNA molecule hybridizes under stringent conditions to SEQ ID NO:10.
62. A method to treat a patient experiencing disease comprising:
- i) characterizing HAS1 isoenzyme variant expression in a cell or cell population;
  - ii) evaluating aberrant HAS1 isoenzyme variant expression; and
  - iii) administering compounds to the cell or cell population resulting in diminished HAS1 isoenzyme variant activity.
63. The method of claim 62 wherein HAS isoenzyme activity is diminished through decreased mRNA translation.
64. The method of claim 62 wherein mRNA translation is decreased through

administration of agents that selectively bind to HAS isoenzyme variant mRNA.

65. The method of claim 64 wherein the agent is anti-sense RNA.

66. The method of claim 64 wherein the agent is anti-sense DNA.

67. The method of claim 64 wherein the agent is small inhibitory RNA.

5 68. The method of claim 62 wherein the HAS1 isoenzyme variant activity is diminished through decreased activity of the HAS1 isoenzyme variant protein.

69. The method of claim 68 wherein protein activity is decreased through administration of an agent which selectively binds to HAS1 isoenzyme variants.

70. The method of claim 69 wherein the agent is a peptide.

10 71. The method of claim 70 wherein the agent is an antibody.

72. The method of claim 70 wherein the agent is an antibody fragment.

73. The method of claim 68 wherein the agent is vesnarinone.

74. The method of claim 68 wherein the agent is hyaluronic acid.

75. A method to treat a patient susceptible to disease comprising:

- 15 i) characterizing HAS1 isoenzyme variant expression in a cell or cell population;  
Evaluation of aberrant HAS1 isoenzyme variant expression; and  
ii) administering at least one compound to the cell or cell population resulting in diminished HAS1 isoenzyme variant activity.

20 76. The method of claim 75 wherein HAS isoenzyme activity is diminished through decreased mRNA translation.

77. The method of claim 76 wherein mRNA translation is decreased through administration of at least one agent that selectively binds to HAS isoenzyme variant



mRNA.

78. The method of claim 77 wherein the agent is anti-sense RNA.

79. The method of claim 77 wherein the agent is anti-sense DNA.

80. The method of claim 77 wherein the agent is small inhibitory RNA.

5 81. The method of claim 75 wherein the HAS1 isoenzyme variant activity is diminished through decreased activity of the HAS1 isoenzyme variant protein.

82. The method of claim 81 wherein protein activity is decreased through administration of an agent which selectively binds to HAS1 isoenzyme variants.

83. The method of claim 82 wherein the agent is a peptide.

10 84. The method of claim 83 wherein the agent is an antibody.

85. The method of claim 83 wherein the agent is an antibody fragment.

86. The method of claim 83 wherein the agent is vesnarinone.

87. The method of claim 83 wherein the agent is hyaluronic acid.

15 88. A method to monitor malignant cells in a human comprising detection of HAS isoenzymes or isoenzyme variants in a sample of cells or cell population from a human.

89. The method of claim 88 wherein the human is suffering from Multiple Myeloma.

90. The method of claim 88 wherein the human is suffering from Waldenstrom's Macroglobulemia.

20 91. A kit for characterizing HAS isoenzyme or isoenzyme variant expression in a cell or cell population comprising:

i) the DNA of claims 51, 54, 57 or 60,

ii) compounds and enzymes sufficient to enable specific fragments of DNA to be

increased in number, and

iii) instructions enabling one to amplify and identify HAS isoenzyme or isoenzyme variant specific fragments.

92. The kit of claim 91 wherein characterizing HAS isoenzyme or isoenzyme variant  
5 expression is used to monitor previously diagnosed Waldenstrom's  
Macroglobulemia.

93. The kit of claim 91 wherein characterizing HAS isoenzyme or isoenzyme variant  
expression is used to monitor previously diagnosed Multiple Myeloma.

94. The kit of claim 91 wherein characterizing HAS isoenzyme or isoenzyme variant  
10 expression is used to diagnose Multiple Myeloma.

95. The kit of claim 91 wherein characterizing HAS isoenzyme or isoenzyme variant  
expression is used to assess clinical outcome of Multiple Myeloma patients.

96. A kit for characterizing HAS isoenzyme variant expression in a cell or cell population  
comprising nucleotides capable of binding selectively to, and thereby distinguishing,  
15 HAS isoenzyme or isoenzyme variant transcripts; compounds sufficient to enable  
formation and identification of complex formed between the nucleotides and HAS  
isoenzyme variant transcripts; and instructions enabling one to identify HAS  
isoenzyme variant expression.

97. The kit of claim 96 wherein characterizing HAS isoenzyme or isoenzyme variant  
20 expression is used to monitor previously diagnosed Waldenstrom's  
Macroglobulemia.

98. The kit of claim 96 wherein characterizing HAS isoenzyme or isoenzyme variant

expression is used to monitor previously diagnosed Multiple Myeloma.

99. The kit of claim 96 wherein characterizing HAS isoenzyme or isoenzyme variant expression is used to diagnose Multiple Myeloma.

100. The kit of claim 96 wherein characterizing HAS isoenzyme or isoenzyme variant expression is used to assess clinical outcome of Multiple Myeloma patients.

101. A kit for characterizing HAS isoenzyme or isoenzyme variant expression in a cell or cell population comprising peptides capable of binding selectively to, and thereby distinguishing, HAS isoenzyme or isoenzyme variant proteins; compounds sufficient to enable formation and identification of complex formed between the complex formed between the peptide and HAS isoenzyme or isoenzyme variant protein; and instructions enabling one to identify HAS isoenzyme or isoenzyme variant proteins.

102. The kit of claim 101 wherein characterizing HAS isoenzyme or isoenzyme variant expression is used to monitor previously diagnosed Waldenstrom's Macroglobulemia.

103. The kit of claim 101 wherein characterizing HAS isoenzyme or isoenzyme variant expression is used to monitor previously diagnosed Multiple Myeloma.

104. The kit of claim 101 wherein characterizing HAS isoenzyme or isoenzyme variant expression is used to diagnose Multiple Myeloma.

105. The kit of claim 101 wherein characterizing HAS isoenzyme or isoenzyme variant expression is used to assess clinical outcome of Multiple Myeloma patients.